PHENOTYPIC DISTANCE OF BULGARIAN AND HUNGARIAN COMMON WINTER WHEAT GENOTYPES

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Abstract

The field experiment was conducted during the period 2020-2023 on an experimental field at Institute of Plant Genetic Resources "K. Malkov" in town of Sadovo, central southern Bulgaria. Five Hungarian, 4 standart varieties and 16 advanced lines winter common wheat were included in the experiment. The following traits were reported: grain yield, test weight, 1000 grains weight and plant heigth. The mathematical processing of the obtained results was carried out by Duncan`s multiple range test, mean, min and max values, CV and standard error, PC and Cluster analysis. Greater diversity was observed in the traits 1000 grains weight, test weight and height, while grain yield was less. The analyzes carried out will also help to select parental forms to create new varieties with the potential for high yield and quality.

Key words: common winter wheat, phenotypic distance, cluster analysis, PC analysis.

INTRODUCTION

Winter common wheat (*Triticum aestivum* L.) is a key component of human nutrition. Increasing global population and continuous urbanisation require an expansion of its production. Approximately one-fifth of human calories in various forms are supplied by wheat, which appears to be an important requirement for production security (Hongjie et al., 2019; Li et al., 2019; Nazarenko et al., 2020). The main goal of modern breeding is to create varieties combining high yield, quality and tolerance to biotic and abiotic stress factors (Uhr, 2015). Valuable hybrid populations have been created through hybridization programs and genetic studies. Trials of the resulting materials in given ecological regions help to create adapted high-yielding winter wheat cultivars (Tayyar, 2010; Muhe & Assefa, 2011; Tsenov et al., 2016; Lozada et al., 2021; Nazarenko et al., 2021). Varietal genotype has an important role in increasing yields and reducing its variability by year depending on growing regions. Microclimate has a significant influence in the expression of genetic possibilities related to grain productivity and quality (Tsenov et al., 2004; Uhr & Samodova, 2020). In this regard, the priority is to obtain high yields by breeding

varieties of common winter wheat that can effectively use the environmental conditions and features of the ecological zone in which it is grown, to resist abiotic and biotic stress factors (Andrusevich et al., 2018). The search for new sources of genetic material for breeding programs on grain yield and quality requires the testing and use of domestic and foreign varieties of common winter wheat. An important component in the increasingly changing climatic stresses on plants is the selection of an appropriate varietal structure to ensure good performance in different ecological regions (Yanchev & Yordanova, 2005; Tsenov et al., 2021). Mannu et al., 2018 found that a good strategy for maintaining the yield potential of common winter wheat under global climate change is to optimize its phenology, which appears to be a major factor in adaptation to a given environment. The major phenological genes, together with the height determining genes, play an important role in breeding wheat lines with reduced lodging-resistant height and higher harvest index and hence high yield potential. The influence of these major adaptation and height genes should be considered in modern wheat breeding programs (Gasperini et al., 2012; Nazarenko et al, 2021; Lozada et al., 2021).

The use of different mathematical approaches such as the application of cluster analysis in combination with the application of principal component analysis (PCA) have been widely applied in research to identify and differentiate different lines and varieties of common winter wheat as well as to reveal the factors underlying existing phenotypic differences. The combination of the two analyses (Cluster Analysis and PCA) are applied in breeding when performing comparative studies on quantitative and qualitative traits between a larger number of lines and varieties. They allow interpretation and evaluation of their phenotypic similarity and dissimilarity depending on the objective of the study. The both analyses complement each other and their simultaneous use (conducting) gives us an in-depth information on the significance of studied traits when grouping the studied materials. These approaches allow us to group the different genotypes according to their similarity in the studied traits into approximately homogeneous clusters, which in turn allows for better selection and good combination in the future construction of an appropriate strategy to guide the selection process (Forkman et al., 2019; Gubatov & Delibaltova, 2020; Cheshkova et al., 2020; Ali et al., 2021; Reckling et al., 2021; Tsenov et al, 2022)

The main purpose of the study was to establish the phenotypic distance of Bulgarian and Hungarian genotypes of common winter wheat for use in their hybridization program.

MATERIALS AND METHODS

The competitive varietal trial was carried out in the experimental field of IPGR "K, Malkov", Sadovo in the period 2020-2023. The trial was carried out in four replications with an experimental plot area of 10 m². In the month of May 2022, there was a strong hailstorm and results were not counted. Twenty-five lines and varieties of common winter wheat were studied, including 4 standards, 5 Hungarian varieties and 16 advanced lines. To characterize the grain of the lines studied, the following parameters were recorded: grain yield (kg/da) plant height (cm), test weight (kg/hl) according to BSS ISO 7971:2000 and 1000 kernels weight (g) by weighing two

samples of 500 grains (BSS ISO 520:2003). The analyses were carried out in the grain quality assessment laboratory. The Mathematical treatment used Duncan's multiple range test (DMRT), mean, minimum and maximum values, CV %. The coefficient of variation was used to define the variation of indicators (Dimova & Marinkov, 1999) as: weak - up to 10%, medium - greater than 10% and less than 20%, strong -above 20%,) and standard error. Principle component and cluster analyses were applied to study the biological and economic traits based on the genotypes included in the experiment.

RESULTS AND DISCUSSIONS

For the period 2020-2023, the average monthly air temperature (Table 1) and precipitation amount (Table 2) were monitored to characterize each growing season. In the first year (2019-2020) less than normal rainfall was recorded in October during seedbed preparation, December, January and during germination, flowering, grain filling and maturing. In February, March and April, the rainfall is higher than normal and supports the picking and spindling. The average monthly temperature in April alone is (-0.4°C) below normal and coincides with crop stem elongation. In November and December, temperatures fell below 5°C and the plants can be said to be dormant. In 2020-2021, the average monthly temperature decreases below 5°C only in January, and a negative deviation from the long-term norm of 0.2°C, 0.5°C and 1.3°C is recorded in November, March and April. Temperatures in the winter months of December, January and March are positive and higher than normal. Below normal rainfall is recorded in November during and after sowing, May and July. In 2021-2022, below normal precipitation is recorded in November, January, March, April, May and July. Insufficient moisture is recorded during important phases of wheat development. Average monthly temperatures in October and March are below normal. In December and January temperatures are below 5°C. Negative average monthly temperatures were not recorded during this growing season (vegetative period). In the last year, the average monthly temperature has been

higher than 5ºC degrees throughout the growing season. Negative deviations are recorded in April and May with -0.189 and -1.84ºC. Moisture deficiency was recorded during seed preparation and sowing in October, December, February and March. There is a trend of rainfall deficiency during sowing preparation and May. Average monthly temperatures are lower than normal in March and April, with deviations of -0.2ºC and no more than -1.3ºC. Negative monthly mean temperatures were recorded during the period.

Table 1. Average monthly air temperature for the period 2020-2023

Years/Months	X	XI	XII	I	II	III	IV	V	VI	VII
Average monthly temperature 2019/2020	14.8	10.7	4.1	2.2	6.1	8.9	11.8	18.2	21.6	24.9
Multi-year values 1931–2000	12.6	6.9	2.1	-4.3	2.4	6.3	12.2	17.5	21.2	23.3
Deviation	2.2	3.8	2.0	6.5	3.7	2.6	-0.4	0.7	0.4	1.6
Average monthly temperature 2020/2021	15.2	6.7	5.6	3.3	5.9	5.8	10.9	18.5	22.3	26.6
Multi-year values 1931–2000	12.6	6.9	2.1	-4.3	2.4	6.3	12.2	17.5	21.2	23.3
Deviation	2.6	-0.2	3.5	7.6	3.5	-0.5	-1.3	1.0	1.1	3.3
Average monthly temperature 2021/2022	11.0	7.8	4.0	3.3	5.0	4.9	13.7	18.5	23.0	25.6
Multi-year values 1931–2000	12.6	6.9	2.1	-4.3	2.4	6.3	12.2	17.5	21.2	23.3
Deviation	-1.6	0.9	1.9	7.6	2.6	-1.5	1.5	0.9	1.8	2.3
Average monthly temperature 2022/2023	13.85	9.57	5.58	5.652	6.045	8.69	12.011	15.659	21.797	27.16
Multi-year values 1931–2000	12.6	6.9	2.1	-4.3	2.4	6.3	12.2	17.5	21.2	23.3
Deviation	1.25	2.67	3.48	9.952	3.645	2.39	-0.189	-1.841	0.597	3.86

Table 2. Amount of precipitation for the period 2020-2023

Years/Months	X	XI	XII	I	II	III	IV	V	VI	VII
Precipitation amount for the month - 2019/2020	14.2	80.7	24.9	2.1	49.4	91.5	93.8	40.1	45.9	38.1
Multi-year values	37.4	47.1	49.7	39.3	30.9	39	42.9	56.8	58.4	46.4
Deviation	-23.2	33.6	-24.8	-37.2	18.5	52.5	50.9	-16.7	-12.5	-8.3
Precipitation amount for the month - 2020/2021	72.7	6.3	55.7	96.4	32.8	42.5	78.5	32.7	60.4	19.9
Multi-year values	37.4	47.1	49.7	39.3	30.9	39	42.9	56.8	58.4	46.4
Deviation	35.3	-40.8	6.0	57.1	1.9	3.5	35.6	-24.1	2.0	-26.5
Precipitation amount for the month 2021/2022	167.9	11.9	96.1	30.3	57.9	22.3	31	39.8	159.7	8.3
Multi-year values	37.4	47.1	49.7	39.3	30.9	39	42.9	56.8	58.4	46.4
Deviation	130.5	-35.2	46.4	-9.0	27.0	-16.7	-11.9	-17.0	101.3	-38.1
Precipitation amount for the month - 2022/2023	1.8	49	47.6	54	1.2	34.8	55.3	80.9	80.2	63.5
Multi-year values	37.4	47.1	49.7	39.3	30.9	39	42.9	56.8	58.4	46.4
Deviation	-35.6	1.9	-2.1	14.7	-29.7	-4.2	12.4	24.1	21.8	17.1

Mean values of the studied traits and their corresponding evidences according to Duncan's multiple comparison test between genotypes are presented in the Table 3. The results show that for all four traits there are significant differences between the tested genotypes. Greater variation was observed in the traits thousand kernels weight, test weight and height, while there was less variation in grain yield. According to the coefficient of variation, plant height was characterized by the highest followed by thousand grains weight and grain yield. It is lowest for test weight. The three higher values are characterized as average in terms of its interpretation, which means that there is a well-defined genetic diversity in the sample of genotypes for the three traits. Grain yield averaged 716.3 kg/da over the three years. It ranged from 644.5 kg/da for the line MX 286/1759 to 864.4 kg/da for the Hungarian variety MV-Nemere. The standard variety Sadovo1 has an average of 673.3 kg/da, which places it in the middle of the sample of varieties and lines. Plant height averaged 97.6 cm over the years of testing. The highest average over the three years was line RU 33/3244 and the lowest was variety Avenue. For the trait thousand kernels weight, the highest value averaged over the years was line RU 134/1370 and the lowest was for variety Avenue. The trait is characterised by an average value of 45.4 g over the period. For the trait test weight, the average was 74.4 kg/hl, with the highest value for the line MX 286/1759 and the lowest for the line RU 135/1456.

Table 3. Duncan's multiple test, means, min., max., CV and standard error of 25 genotypes for 4 quantitative traits

Genotype	Yield	Sig	Plant Heigh	Sig	TKW	Sig	Test Weight	Sig
MX 270/ 28	735.4	abc	95.0	fghij	46.6	efghi	74.8	bcdefgh
MX 270/ 50	787.9	bc	96.0	ghij	49.0	ghij	74.2	bcdef
РУ 129/3053	779.2	abc	99.0	ij	44.1	bcdef	76.5	efgh
РУ 33/3244	699.1	ab	100.7	j	48.0	fghij	77.1	fgh
MX 270/ 3461	681.2	ab	94.0	fghij	49.3	ghij	77.2	fgh
MX 285/1058	752.2	abc	98.3	hij	41.5	bc	75.7	cdefgh
РУ 48/2553	687.4	ab	88.7	cdefghi	41.8	bcd	74.9	bcdefgh
MX 286/1759	644.5	a	88.3	cdefgh	46.3	efghi	79.0	h
MX 286/1777	673.2	ab	88.0	cdefgh	43.0	bcde	77.6	fgh
Avenue	660.7	ab	68.7	a	35.8	a	73.8	abcdef
Anapurna	649.6	a	76.7	ab	40.7	b	75.4	cdefgh
Sadovo 1	673.3	ab	94.7	fghij	50.4	hij	76.0	defgh
Enola	653.9	ab	90.0	defghi	44.0	bcdef	78.7	gh
MX 272/3872	655.8	ab	94.7	fghij	50.4	ij	70.8	ab
MX 215/3	668.7	ab	84.3	bcdef	45.8	cdefg	74.3	bcdef
РУ134/1343	689.6	ab	80.7	bcd	46.8	efghi	70.9	ab
РУ177/486	754.7	abc	81.0	bcd	48.4	fghij	74.6	bcdefg
РУ135/1456	695.2	ab	80.7	bcd	47.9	fghij	69.8	a
РУ179/1400	766.6	abc	85.7	bcdefg	47.9	fghij	73.9	abcdef
РУ134/1370	761.4	abc	78.7	bc	51.9	j	71.6	abc
MV-Nador	766.5	abc	76.7	ab	43.4	bcde	72.1	abcd
MV-Nemere	864.4	c	83.0	bcde	40.4	b	72.8	abcde
MV-Menrot	717.3	ab	87.7	cdefg	43.1	bcde	72.5	abcde
MV-Mente	726.1	ab	92.3	efghi	46.0	defgh	73.5	abcdef
MV-Kaplar	762.8	abc	85.7	bcdefg	41.8	bcd	71.9	abcd
mean	716.3	-	87.6	-	45.4 ±	-	74.4	-
± m	± 11	-	± 1.6	-	0.77	-	± 0.5	-
min.	644.5	-	68.7	-	35.8	-	69.8	-
max.	864.4	-	100.7	-	51.9	-	79.0	-
CV%	7.71	-	9.23	-	8.47	-	3.33	-

Mean values (in each column) followed by the same letters (in the subscript) are not significantly different at p < 0.05 according to Duncan's multiple range test (DMRT)

PC analysis was applied to study the biological and economic traits based on the genotypes included in Table 3. Figure 1 shows the PC analysis for the four traits studied. As can be seen from the figure, more than 70% of the total variation in phenotypic expression of genotypes across traits is due to the first two principal components, PC1 and PC2. This value is large and correct discussion of the results is possible. In the figure, grain yield (Y) is negatively correlated with test weight (TW) and plant height (H) and weakly positively correlated with thousand kernels weight (TKW). Thousand kernels weight is positively correlated with plant height and negatively correlated with test weight. Plant height is strongly positively correlated with thousand kernels weight and weakly positively correlated with test weight and negatively correlated with grain yield.

The locations of the common winter wheat genotypes from the CVT relative to the principal components PC1 and PC2 are presented in Figure 2. By comparing the two figures (Figure 1 and Figure 2) we can assess which genotypes are more strongly associated with which traits. It is noteworthy that most of the genotypes studied are associated with yield. It is noticeable that the individual varieties and lines are located in the middle of the coordinate system. This suggests that they are balanced with respect to the traits tested. Genotypes located at the periphery indicate that they have superiority in any of the traits tested. Impressive are the new lines, which are located around the center of the figure and should behave in a balanced manner in most respects.
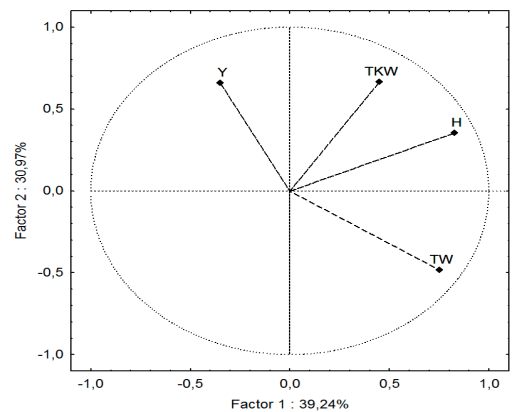

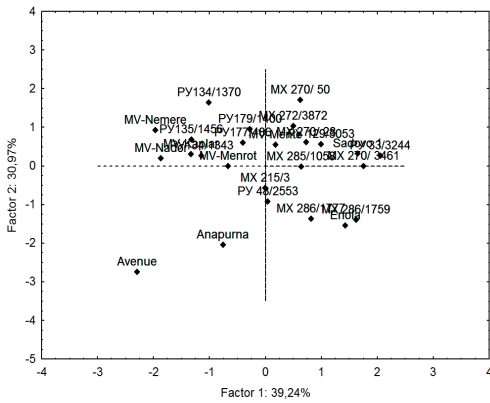Figure 1. PC analysis of yield, 1000 kernels weigh, plant height and test weight

Figure 2. PC analysis of 25 common winter wheat genotypes

By comparing the two figures (Figure 1 and Figure 2) we can assess which genotypes are more strongly associated with which traits. It is noteworthy that a large proportion of the genotypes examined are associated with yield. This is where the lines suitable for presentation as new varieties are located. They are suitable for inclusion in the hybridisation scheme, as they would contribute to increasing productivity. The two standards, Avenue and Annapurna, occupy a neutral position in the coordinate system and are not characterized by any of the traits studied. It is noticeable that the individual varieties and lines are located in the middle of the coordinate system. This suggests that they are balanced with respect to the traits tested. The genotypes located at the periphery indicate that they have superiority in any of the traits. The new breeding lines that lie around the centre of the figure are impressive and should be more balanced on most traits.

The dendrogram shows that two major clusters with genetic distance are formed (Figure 3). The advanced lines are located in both clusters, subsequently splitting into two subclusters. In the leftmost cluster, newly established lines with RU 134/1370, RU 179/1400, RU 177/486, and RU 135/1456 and RU 134/1343 and MX 272/3872 are located, and in the second subcluster of the first cluster, foreign varieties are located. The right cluster has two smaller clusters. The left subcluster has the two Bulgarian standards and three newly established lines. This defines the lines as very close to the Bulgarian standards. In the right sub-cluster of cluster two are located for the

most part lines of origin MX 270/28 and MX 270/50. For smaller and faster breeding progress we should cross genotypes located in the same cluster and vice versa for larger but slower progress cross genotypes from genetically distant clusters. According to this analysis, which gives a good idea of the genetic proximity and distance of the breeding materials, an appropriate strategy can be built to guide the breeding process. It is recommended to cross the Enola and Sadovo1 standards with genotypes from the other cluster to achieve higher but slower results.
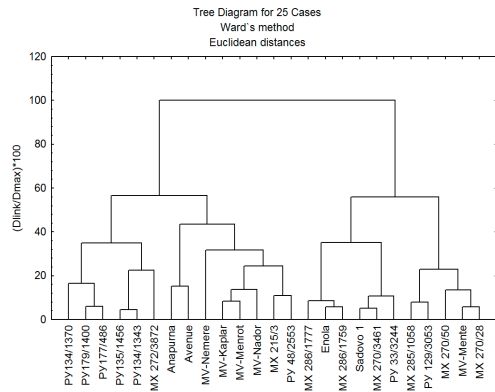

Figure 3. Cluster analysis of 25 common winter wheat genotypes

CONCLUSIONS

In the meteorological surveys, a trend of rainfall deficiency emerged during the preparation of sowing and the month of May. Monthly average temperatures are lower than normal in March and April, with deviations of -0.2ºC and no more than -1.3ºC. No negative monthly mean temperatures were recorded during the period.

The presented mean values of the tested traits and their corresponding evidences according to Duncan's multiple comparison test between genotypes show that for all four traits there are significant differences between the tested genotypes.

In the correlation analysis, grain yield was negatively correlated with test weight and plant height and weakly positive correlated with thousand kernels weight. Thousand kernels weight is positively correlated with plant height and negatively correlated with test weight.

Plant height is strongly positively correlated with thousand kernels weight and weakly positive correlated with test weight and negatively correlated with grain yield.

The PCA results show that more than 70% of the total variation in the phenotypic expression of the genotypes for the traits is due to the first two main components - PC1 and PC2 a large part of the studied genotypes are related to yield. This suggests that they are balanced with respect to the traits tested.

Cluster analysis, gives a good idea of the genetic proximity and remoteness of the breeding materials, an appropriate strategy can be built to guide the breeding process. It is recommended to cross the standards Enola and Sadovo1 with genotypes from the other cluster to achieve higher but slower results.

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